

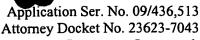
Application Ser. No. 09/436,513 Attorney Docket No. 23623-7043

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Appendix I - Clean Copy of Pending Claims

- 1. (Amended) A modified serine hydrolase, said hydrolase comprising an amino acid residue in a subsite replaced with a cysteine, wherein the cysteine is modified by replacing the thiol hydrogen in the cysteine with a substituent group providing a thiol side chain comprising a chiral substituent.
- 2. The modified serine hydrolase of claim 1, wherein the serine hydrolase catalyzes a transamidation.
- 3. The modified serine hydrolase of claim 1, wherein the serine hydrolase catalyzes a transpeptidation.
- 4. The modified serine hydrolase of claim 1, wherein the serine hydrolase catalyzes a transesterification.
- 5. The modified serine hydrolase of claim 1, wherein said serine hydrolase is selected from the group consisting of an alpha/beta serine hydrolase, a subtilisin type serine protease, and a chymotrypsin serine protease.
- 6. The modified serine hydrolase of claim 1, wherein said serine hydrolase is a subtilisin.
- 7. The modified serine hydrolase of claim 6, wherein said serine hydrolase catalyzes a transamidation and is stereoselective.
- 8. The modified serine hydrolase of claim 6, wherein the amino acid replaced with a cysteine is an amino acid in the S_1 , S_1 , or S_2 subsite.
- 9. The modified serine hydrolase of claim 8, wherein the amino acid replaced with a cysteine is selected from the group consisting of asparagine, leucine, methionine, and serine.
- 10. (Amended) The modified serine hydrolase of claim 8, wherein said amino acid is selected from the group consisting of amino acid 156 in the S_1 subsite, amino acid 166 in the S_1 subsite, amino acid 217 in the S_1 ' subsite, amino acid 222 in S_1 ' subsite and amino acid 62 in the S_2 subsite.
- 11. (Amended) The modified serine hydrolase of claim 8, wherein said substituent is selected from the group consisting of an enantiomerically pure oxazolidinone, an enantiomerically pure indenone, and an enantiomerically pure phenyl-ethyl-thiol.



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phenyl-2-oxazolidinone, N-(3'-thio-propyl)-(S)-4-phenyl-2-oxazolidinone, N-(3'-thio-propyl)-(R)-4-benzyl-2-oxazolidinone, N-(3'-thio-propyl)-(S)-4-benzyl-2-oxazolidinone, N-(2'-thio-ethyl)-(R)-4-phenyl-2-oxazolidinone, N-(2'-thio-ethyl)-(S)-4-phenyl-2-oxazolidinone, N-(2'-thio-ethyl)-(S)-4-benzyl-2-oxazolidinone, N-(2'-thio-ethyl)-(S)-4-benzyl-2-oxazolidinone, N-(3'-thio)-(3aR-cis)-3,3a,8,8a-tetrahydro-2H-indeno[1,2-d]-oxazol-2-one, and N-(3'-thio)-(3aS-cis)-3,3a,8,8a-tetrahydro-2H-indeno[1,2-d]-oxazol-2-one.

51. (Amended) A method of producing a chemically modified mutated serine hydrolase, said method comprising

providing a serine hydrolase, said hydrolase comprising an amino acid residue in a subsite replaced with a cysteine; and

modifying the cysteine by replacing the thiol hydrogen in the cysteine with a substituent group providing a thiol side chain comprising a chiral substituent.

- 52. The method of claim 51, wherein said hydrolase is selected from the group consisting of an alpha/beta serine protease, a subtilisin type serine protease, and a chymotrypsin serine protease.
 - 53. The method of claim 51, wherein said hydrolase is a subtilisin.
- 54. The method of claim 53, wherein the amino acid replaced with a cysteine is an amino acid in the S₁, S₁', or S₂ subsite.
- 55. The method of claim 53, wherein the amino acid replaced with a cysteine is selected from the group consisting of asparagine, leucine, methionine, and serine.
- 56. (Amended) The method of claim 53, wherein said amino acid is selected from the group consisting of amino acid 156 in the S_1 subsite, amino acid 166 in the S_1 subsite. amino acid 217 in the S_1 ' subsite, amino acid 222 in S_1 ' subsite and amino acid 62 in the S_2 subsite.
- 57. (Amended) The method of claim 53, wherein said substituent is selected from the group consisting of a chiral oxazolidinone, a chiral indenone, and a chiral phenyl-ethyl-thiol.
- 59. (Amended) The method of claim 53, wherein said substituent is selected from the group consisting of (R)-2-methoxy-2-phenyl-ethyl-thiol, (S)-2-methoxy-2-phenyl-ethyl-thiol, (R)-2-hydroxy-2-phenyl-ethyl-thiol, (S)-2-hydroxy-2-phenyl-ethyl-thiol, N-(3'-thio-propyl)-2-oxazolidinone, N-(3'-thio-propyl)-(R)-4-isopropyl-2-oxazolidinone, N-(3'-thio-propyl)-(R)-4-phenyl-2-oxazolidinone, N-(3'-thio-propyl)-(R)-4-benzyl-2-oxazolidinone, N-(3'-thio-propyl)-(R)-4-benzyl-2-oxazolidinone, N-(2'-thio-ethyl)-(S)-4-benzyl-2-oxazolidinone, N-(2'-thio-ethyl)-(R)-4-benzyl-2-oxazolidinone, N-(2'-thio-ethyl)-(S)-4-benzyl-2-oxazolidinone, N-(2'-thio-ethyl)-(R)-4-benzyl-2-oxazolidinone, N-(2'-thio-ethyl)-(S)-4-benzyl-2-oxazolidinone, N-(3'-thio)-(3aR-cis)-3,3a,8,8a-tetrahydro-2H-indeno[1,2-d]-oxazol-2-one, and N-(3'-thio)-(3aS-cis)-3,3a,8,8a-tetrahydro-2H-



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indeno[1,2-d]-oxazol-2-one.

61. The method of claim 53, wherein said method further comprises screening the modified serine hydrolase for an activity selected from the group consisting of a transesterification activity, a transamidation activity, and a transpeptidation activity.

62. The method of claim 61, wherein said activity is stereoselective.